

# UBE2D4 (UbcH5d) [untagged]

## E2 – Ubiquitin Conjugating Enzyme

Alternate Name: LOC51619 protein, UbcH5d

Cat. No. **62-0017-020**  
Lot. No. **30020**

Quantity: 20 µg  
Storage: -70°C

FOR RESEARCH USE ONLY

NOT FOR USE IN HUMANS



### CERTIFICATE OF ANALYSIS

## Background

The enzymes of the ubiquitylation pathway play a pivotal role in a number of cellular processes including regulated and targeted proteasomal degradation of substrate proteins. Three classes of enzymes are involved in the process of ubiquitylation; activating enzymes (E1s), conjugating enzymes (E2s) and protein ligases (E3s). UBE2D4 is a member of the E2 ubiquitin-conjugating enzyme family and the human gene was first described by Colland *et al.* (2004).

### References:

Colland F, Jacq X, Trouplin V, Mouglin C, Groizeleau C, Hamburger A, Meil A, Wojcik A, Legrain P, Gauthier J (2004) Functional proteomics mapping of a human signaling pathway. *Genome Res* 14, 1324-32.

## Physical Characteristics

**Species:** human

**Source:** *E. coli* expression

**Quantity:** 20 µg

**Concentration:** 1 mg/ml

**Formulation:** 50 mM HEPES pH 7.5,  
150 mM sodium chloride, 2 mM  
dithiothreitol, 10% glycerol

**Molecular Weight:** ~20 kDa

**Purity:** >98% by InstantBlue™ SDS-PAGE

**Stability/Storage:** 12 months at -70°C;  
aliquot as required

### Protein Sequence:

GSHMASMTGGQQMGRGSEFELGSTSNGRQCA  
GIRPCAAAM***ALKRIQKELTDLQRDPPAQCSAG***  
PVGDDLFWQATIMGPNDSPYQGGVFFLT  
HFPTDYPFKPKVAFTTKIYHPNINSNGSI  
CLDILRSQWSPALTVSKVLLSICSLLCDPNPD  
DPLVPEIAHTYKADREKYNRLAREWTQKYAM

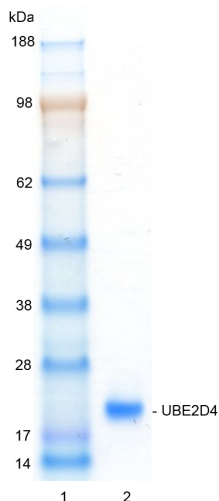
The residues underlined remain after cleavage and removal of the purification tag.

UBE2D4 (regular text): Start ***bold italics*** (amino acid residues 1-147)

Accession number: NP\_057067

## Quality Assurance

**Purity:**  
4-12% gradient SDS-PAGE  
InstantBlue™ staining  
Lane 1: MW markers  
Lane 2: 1 µg UBE2D4



### Protein Identification:

Confirmed by mass spectrometry.

### E2-Ubiquitin Thioester Loading Assay:

The activity of UBE2D4 was validated by loading E1 UBE1 activated ubiquitin onto the active cysteine of the UBE2D4 E2 enzyme via a transthioylation reaction. Incubation of the UBE1 and UBE2D4 enzymes in the presence of ubiquitin and ATP at 30°C was compared at two time points, T<sub>0</sub> and T<sub>10</sub> minutes. Sensitivity of the ubiquitin/UBE2D4 thioester bond to the reducing agent DTT was confirmed.



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Lot-specific COA version tracker: v1.0.0